Development and Formulation of Millet based Nutri-Bar

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Abstract

Whole grains like millet and rice are rich in nutrients and fiber, making them an excellent ingredient for developing a nutritious snack bar. The nutri bar was formulated using popped pearl millet and black rice flakes as primary ingredient. Three samples (F1, F2 and F3) with different composition are prepared. The proximate composition, chemical and sensory analysis was determined for all the samples. Sensory evaluation was conducted using a nine-point hedonic scale to assess the bar's taste, colour, texture, and overall acceptability. Results showed that the sample F3 has rich content of 12.8 gm of protein, 77.6 gm of carbohydrate, 366 Kcal of calories, 9.4 gm of dietary fiber were recorded per 100 grams of sample. Hence sample F3 is considered as the perfect formulation as it scored 8.5/9 in hedonic scale.

Keywords: *popped pearl millet, black rice flakes, sensory, nutrition, snack bar.*

1. Introduction

Considering the significant increase in the popularity of (RTE –Ready to Eat) snack options like nutrition bars as a quick and easy snack due to the global demand for convenient and healthy food options. In the view of formulating an innovating nutrition bar, millet and rice infusion plays important role in food market. Whole grains like millet and rice are rich in essential nutrients, fiber and antioxidants making them ideal for coping up with malnutrition for every individual. This study focuses on development and formulation of nutrition bar with pearl millet and black rice. Using popped millet and flaked rice as primary ingredient offers a novel formulation of snack bar. The findings of studies has shown that utilizing popped pearl millet contributes to the improvement of nutritional value by inactivating some of the anti-nutritional factors (enzymes and enzyme inhibitors) and thereby enhancing the protein and carbohydrate digestibility [1]; it also enhances the appearance, colour, taste and aroma of the processed raw material. the findings of studies also shows that flaking of black rice (also known as forbidden rice) by using traditional method increases the digestibility, overall acceptability and reduces water activity when added to bar [3]. Therefore this paper aims on formulating well acceptable nutrition bar with best suitable processing method and evaluation of its nutritional value and psycho-chemical properties.

2. Materials and Methods

2.1 Selection of Raw Materials

In the present study ingredients such as pearl millet, flaked black rice, dates and jaggery were selected to prepare the nutrition bar. Millet was produced from the local market of Coimbatore, Tamilnadu because of its easy proximity. Other ingredients such as flaked black rice, dates and jaggery were purchased from nearby departmental store. The ingredients were cleaned and foreign particles were removed and stored in proper containers.

2.2 Processing of Raw Materials

The whole grain millet of known weight is taken and conditioned to obtain uniform moisture content of 16%. Then the conditioned pearl millet is added to hot salt bed in the ratio of 1:20 (by weight) which is maintained at 260°C. Maximum amount of millets are popped within 15 seconds [5]. By using 12 mesh size sieve the popped millets are separated from salt. The method of using salt bed is adopted because of its high yield of popping when compared to other methods. Other ingredients like flaked black rice is readily purchased from local markets and coarsely grinded using food processor to improve the binding, texture and overall acceptability of product. Seeds are removed from dates and cut to small pieces.

2.3 Preparation of Bar

Initially for the preparation of bar, boil the jaggery for few minutes in medium flame until it reaches the hard crack stage (150-155°C). Now toss in all prepared dry ingredients into the jaggery when it is hot. To improve the binding property of product slightly pulse the popped millet and black rice flakes using a food processor; finally add diced dates into it and give a good mix using spatula or ladle. After mixing immediately transfer the mix in to tray with butter sheet; with the help of roller gently spread the mixture into 12×12 cm square with the thickness of 1.5 to 2 cm. leave it to cool down and cut into rectangle shaped bars and pack it with either cling film or aluminum foil and store it in a air tight container.

Table 1.Composition of Ingredients

Ingredients	Formula		
	F1	F2	F3
Popped pearl millet (g)	30	35	40
Black rice flakes (g)	20	20	20
Dates (g)	25	15	10
Jaggery (g)	25	30	30

Figure1. Picture of F3 Bar

3. Nutritional Analysis of the Bar

3.1 Determination of Fat Content.

Reagents: Petroleum Ether -- of boiling range 40°C to 60°C.

Procedure: Weigh accurately about 2.5 g of the dried material into thimble and extract with petroleum ether in a Sox let or other suitable extractor. The extraction period may vary from 4 hours at a condensation rate of 5 to 6 drops per second to 16 hours at 2 to 3drops per second. Dry the extract on a steam-bath for 30 minutes, cool in a desiccator and weigh. Continue at 30 minute intervals this alternate drying and weighing until the difference between two successive weighing is less than one mg. Note the lowest mass.

Calculation: Crude fat (on moisture-free basis) = $\frac{100\ (M_1 - M_2)}{m}$ % by mass

Where

M_1 = mass in 'g' of the extraction flask with dried extract,
M_2 = mass in 'g' of the extraction flask, and
m = mass in 'g' of the dried sample taken for the test

Reference: AOAC 18th Edition / FSSAI

3.2 Determination of Protein Content

Principle: The protein content is determined from the organic Nitrogen content by Kjeldahl method. The various nitrogenous compounds are converted into ammonium sulphate by boiling with concentrated sulphuric acid. The ammonium sulphate formed is decomposed with an alkali (NaOH) and the ammonia liberated is absorbed in excess of standard solution of acid and then back titrated with standard alkali.

Apparatus:

a. **Kjeldahl digestion flask** - 500 or 800 ml
b. **Kjeldahl distillation apparatus,** - same digestion flask fitted with rubber stopper through which passes lower end of efficient rubber bulb or trap to prevent mechanical carryover of NaOH during distillation or apparatus as shown below.
c. **Conical flask, 250 ml**
d. **Burette 50 ml**.

Reagents:

a. **Concentrated Sulphuric acid** - sp gr 1.84
b. **Sodium Hydroxide solution** - 45%. Dissolve 450 gm of Sodium Hydroxide in 1000 ml water
c. **Standard Sulphuric acid solution** – 0.1 N

d. **Standard Sodium Hydroxide solution** – 0.1
e. **Methyl red Indicator solution** - Dissolve 0.5 gm methyl red in 100 ml of alcohol

Procedure: Weigh quickly about 1-2 g of the sample and transfer to a 500 or 800 ml Kjeldahl flask taking care to see that no portion of the sample clings to the neck of the flask. Add 0.7 gm. of Mercuric oxide, 15 gm. of Potassium Sulphate and 40 ml of concentrated sulphuric acid (Mercuric oxide is added to increase the rate of organic breakdown during acid digestion. Because of environmental/safety concerns over handling and disposal of mercury, copper sulphate can be used. This is important from safety point of view as mercury vapors might escape into the environment during the distillation process. Also Missouri catalyst tablets known as Kjeldahl tablets (Composition: 48.8% Sodium sulphate & 48.9% Potassium sulphate & 0.3 % copper sulphate can also be used). Add two to three glass beads. Place the flask in an inclined position on the stand in the digestion chamber and digest. Heat the flask gently at low flame until the initial frothing ceases and the mixture boils steadily at a moderate rate. During heating rotate the flask several times. Continue heating for about an hour or more until the colour of the digest is pale blue. If Black specs are present after 30 minis of digestion, wrap the vessel with aluminum foil and keep for 2-3 minis. By doing this black specs would move down from the walls in the digestion mixture. If the specs are still present, remove the vessel from heat and allow to cool for 10 mins. Do not modify the heat intensity in the whole process. Alternatively, few drops of water may also be pour down across the side of the flask. Cool the digest and add slowly 200 ml of water. Cool, add a piece of granulated Zinc or anti bump granules and carefully pour down the side of the flask sufficient NaOH solution (450 gm/liter) to make the contents strongly alkaline (about 110 ml) before mixing the acid and alkaline layer.

Connect the flask to a distillation apparatus incorporating an efficient flash head and condenser. To the condenser fit a delivery tube which dips just below the surface of the pipette volume of standard acid contained in a conical flask receiver. (Precaution: The receiving solution must remain below 45 ^{o}C to prevent loss of ammonia). Mix the contents of the digestion flask and boil until 150 ml have distilled into the receiver. Add 5 drops of methyl red indicator and titrate with 0.1 N NaOH solutions.

Carry out a blank titration simultaneously.1 ml of 0 .1 N (H_2 SO_4) = 0.0014gm N.

Calculations:

$$\text{Nitrogen content (N) in \%} = \frac{(\text{Blank} - \text{Titre value}) \times \text{Normality} \times 1.4}{\text{Sample Weight}}$$

Calculate protein % : N x Convertion factor

Ideally the protein content of food stuff is calculated by multiplying its total nitrogen content by 6.25, This factor is used whenever the nature of the protein is unknown or when the product to be analyzed is a mixture of different proteins with different factors. However use of different Nitrogen conversion factors for different matrices may lead to better accuracy of results.

Reference: FSSAI Manual of methods

3.3 Determination of Total Carbohydrate by Calculation Method

c The carbohydrate content of the formulated bar samples were determined as the nitrogen – free extraction calculated by differences as described by. This was done by subtracting the sum of protein, crude fat, moisture, crude fibre and ash from 100. Percentage (%) carbohydrate = 100 – (protein + crude fat + moisture + crude fibre + ash) %.

Calculations: Carbohydrate % : 100 –(Fat + Protein + Ash + Moisture)

3.4 Determination of Energy/Calories

Principle: The energy value of the fruit bars were calculated using the protein, fat and carbohydrate contents according to the method described by AOAC

Calculations: (Total Fat value x 9) + (Protein value x 4) + (Carbohydrate value x 4) = Energy

3.5 Estimation of Sugar

Reagents:

a. **Fehling's Solution -1** – 34.6g of pure copper sulfate was dissolved in distilled water and the solution was made up to 500 ml.
b. **Fehling's Solution -2**- 70 g Sodium hydroxide with 173 g Rochelle salt (sodium potassium tartrate) were taken and dissolved in distilled water and made the solution up to 500 ml with water,
c. **Methylene Blue (1%):** 0.5 g methylene blue was taken and volume was made 50 ml,
d. **Neutral lead acetate solution (45**%): 225 g of neutral lead acetate was dissolved in distilled water and the solution was made up to 500 ml.
e. **Sodium Oxalate solution**: 22g of sodium oxalate was dissolved in distilled water and the volume was made 100 ml.
f. **Standard invert sugar solution:** 9.5 g of sucrose was taken onto a 1- litre volumetric flask. 100 ml water and 5 ml con HCl was added. It was allowed to strand for 3 days 20°-25 c

Estimation of Total Sugar: 5g of the sample was taken in a 500 ml beaker. 100 ml warm water was added and neutralized with 10% NaOH. 2 ml of lead acetate solution was added and it was stand for 10 min. the necessary amount of sodium oxalate solution was added to remove the excess of lead. The volume was made up to remove the excess of lead. The volume was made to 250 ml with distilled water and filtered. 50 ml of the clarified and deleaded solution was transferred solution was transferred to a 250ml flask. 10 ml 1N HCl was added into the flask. This solution was then boiled for 2 min. After cooling, 2-3 drops of phenolphthalein was added and the contents were neutralized with NaOH. The solution was filtered and the volume was made 250 ml. Thus the sample solution was prepared. 10 ml of a mixed Fehling's solution (5 ml Fehling's solution-1 and 5 ml Fehling's -2) were pipette into a conical flask. A burette was filled with the clarified sample solution and running the whole volume required reduce the Fehling's solution so that, 0.5 -

1.0 ml was still required to complete the titration. The content of the flask was mixed and then heated to boiling for 2 min. Three drops of methylene blue indicator were added. Then the titration continued till color completely disappeared.

Calculations: % Total Sugar = mg /100ml x Dilution x 100 Wt of the samplex1000

3.6 Determination of Total Dietary Fiber

Procedure: Blank was run along with samples to measure any contribution from reagents to residue. Defatting of samples have done with 25ml of petroleum ether / g of sample three times to remove fixed oil. Then, Weigh 1gm of sample. Phosphate buffer (pH 6.0, 50 mL) was added to sample. Adjust to pH 6.0 ± 0.2 by adding 0.3N NaOH or 0.3N HCl. Enzyme hydrolysis of sample was started by adding 0.1mL α-amylase, incubate at 95 – 100 °C for 30 minutes in water bath with continuous agitation. Cool to room temperature. Adjust to pH 7.5 ± 0.2 by adding 10ml of 0.3N NaOH. Papain 5mg was added, incubate at 60 °C for 30 minutes in water bath with continuous agitation. Cool to room temperature. The pH was adjusted to 4.0 – 4.6 by adding 10ml 0.3N HCl. Amyloglucosidase 0.3mL was added, incubate at 60 °C for 30 minutes let precipitates to form and filter it. Weigh the residue.

Estimate soluble dietary fibers: filtrates plus washing were mixed with 400mL of 95% ethanol to precipitate materials that were soluble in the digestates. After 1h, precipitates were filtered. Residue was washed successively three times with 20mL of 78% ethanol and two times with 10mL of 95% ethanol and then acetone respectively.

For insoluble dietary fiber estimation: residue was washed with 10mL of water (for removing soluble dietary fibers), 95% ethanol and then acetone respectively. Residue was dried at 105°C for 5h in hot air oven, cool in dessicator and weigh to 0.1mg separately (S1 and S2).

The soluble and insoluble dietary fibers (%) were calculated by using following formula.

$$\text{Sol.DF } \% = \frac{(\text{Residue (S1)- Protein-Ash – Blank})}{\text{Weight of test portion}} \times 100$$

$$\text{Insol.DF } \% = \frac{(\text{Residue (S2)- Protein-Ash – Blank})}{\text{Weight of test portion}} \times 100$$

Total dietary fiber % = Soluble DF + Insoluble DF

Reference: The raw samples were analyzed by Prosky method for IDF and SDF according to AOAC Method 993.19 and 991.42, an enzymatic-gravimetric

4. Chemical Analysis of the Bar

4.1 Determination of Water Activity:

Hygrometric methods Salt method: The aw level may be determined relatively cheaply using apparatus with the salt/filterpaper method of Kvaale and Dalhoff (1963), as modified by Northolt and Heuvelman (1982) and Hilsheimer and Hauschild (1985).
The method is based on the fact that dried salt adhering to filter paper is dissolved if the humidity of the surrounding air has reached a point equal to the saturation humidity of the salt. If salts of different saturation humidities are used, the aw level of a sample may be estimated.

Thread hygrometers: This method is based on the hygroscopicity of the polyamide thread, which reacts to an increase in moisture by elongating noticeably. The instrument consists of a sample-scale pan and an attachment that is connected from the pan to a measuring unit by means of a lever system. The polyamide thread is inside the attachment. The change in thread length is converted to a scale on an indicator by means of an axle and lever mechanism. Measuring water activity with this instrument (which takes about 3–4 hours) requires constant temperature.

5. Microbiological Analysis of the Bar

5.1 Total Microbial Count

Reference: IS 5402:2012/ISO 4833:2003,BAM,*DGHS Manual* (2005)

Principle: Two poured plates are prepared using a specified culture medium with specified quantity of sample (if liquid) or Initial suspension (If solid) & other pair of plates prepared under same condition with decimal dilution of test sample incubated at **35°C for 48 ± 2 hrs**

Diluents: 0.1% Peptone salt solution – Himedia M1748 Sterilized by autoclaving at 121° C/15 lbs pressure.

Media Preparation: Himedia MO91 - pH 7.0±0.2
Suspend 2.35 grams in 100 ml distilled water. Heat to boiling to dissolve the medium completely Sterilize by autoclaving at15 lbs pressure (121°C) for 15 minutes.
Cool in water bath at 44 °C to 47 °C before use, Mix well before pour into sterile Petri plates.

Procedure:

- Take two sterile Petri dishes transfer to each dish, by means of a sterile pipette 1 ml of thetest sample (liquid)/ 1 ml of the initial suspension (Solid/Semisolid) ($10^{\square}$dilution).
- Take two other sterile Petri dishes transfer to each dish, by changing the tip 1 ml of thetest sample $10^{\square}$dilution for solid /semisolid , 10^{2} dilution for liquid .
- Fallow similar procedure until the required dilution to be plated
- Pour about 12 ml to 15 ml of the plate count agar (5.2) at 44 °C to 47 °C into each Petri dish;carefully mix the inoculum with the medium by rotating the Petri dishes.
- Allow the mixture to solidify by leaving the Petri dishes standing on a cool horizontal surface
- If suspected over growth over lay 4ml of medium

Method of Calculation: 10 to 250 Colonies

The Number *N* of microorganisms present in the test sample per ml (liquid products) or per gram (other Products).

$$N = \frac{\sum C}{[(1 \times n_1) + (0.1 \times n_2) \times (d)}$$

where:
N = Number of colonies per ml or g of product
∑ C = Sum of all colonies on all plates counted
n_1 = Number of plates in first dilution counted
n_2 = Number of plates in second dilution counted
d = Dilution from which the first counts were obtained

6. Sensory Evaluation of the Bar

To assess the consumer acceptability of the prepared millet based nutria-bar, a sensory evaluation was conducted. This evaluation involved a trained panel of 8 judges who assessed the bar based on its sensory characteristics. The organoleptic study, which focuses on the sensory properties of the product, was carried out over a period of approximately 2 months. The judges evaluated the millet bar for the following sensory attributes which includes colour, texture, flavour, overall acceptability. The sensory evaluation employed a 9-point hedonic scale developed by Larmand. This type of scale allows judges to rate their level of liking or disliking for a product on a numbered scale, typically ranging from "dislike extremely" (low score) to "like extremely" (high score).

Table 2.Sensory Evaluation of Bar with Different Composition

Panelist	Colour			Texture			Aroma			Taste		
	F1	F2	F3	F1	F2	F3	F1	F2	F3	F1	F2	F3
1	5	7	8	6	7	7	9	9	9	6	8	9
2	6	7	9	7	8	9	9	9	8	7	7	9
3	7	7	7	4	6	9	9	9	8	8	7	8
4	5	9	8	6	9	8	9	7	9	8	9	9
5	7	8	8	8	7	9	9	8	9	7	8	8
6	8	9	9	6	8	7	8	9	7	7	8	9
7	5	6	7	7	8	8	9	9	9	8	9	9
8	6	9	9	4	8	9	9	9	9	6	8	9

The table 2 shows the overall acceptability for sample F3 was high when compared to other samples F1 and F2. F3 scored 8.5/9 in overall acceptability. Hence the colour,

texture, aroma and taste for F3 was extremely likable by the panelist members. And so sample F3 is taken as the final composition.

7. Results and Discussion

7.1 Proximate Composition of the Bar

Table 3 shows the proximate compositon of the millet bar samples. The results were provided in terms of grams per 100 g of sample. Test like fat, protein, carbohydrate, dietary fiber, calories and total sugar were taken for all three samples. The level of fat and carbohydrate on each samle were similar and are within accepatable range. All the three samples have good amount of protein around 13g per 100 g, which is enough according to RDA. It is evident from the result that the total suagr content increases as the concentration of dates and jaggery increases. The total dietary fiber ranged from 9.4 g in F1, 9.8 g in F2 and 9.6 g in F3 there were no significant changes between samples.

Table 3.Proximate Composition of millet-rice bar samples

Sample	Fat (g)	Protein (g)	Carbohydrates (g)	Calories (Kcal)	Total Sugar (g)	Dietary Fiber (g)
F1	0.6	13.1	76.2	36I	49.8	10.3
F2	0.6	12.9	79.4	359	48.1	9.6
F3	0.5	12.8	77.6	366	47.2	9.4

7.2 Water Activity (aw) test

For controlling the shelf-life of food products, water activity is an important mean. The water activity of breakfast cereal increased significantly ($P \leq 0.05$) from 0.17 to 0.40 (approximately 42% increase).The test of measuring the availability of water for biological reaction is called water activity test. It determines the ability of micro-organisms to grow. If water activity decreases, micro-organisms with the ability to grow will also decrease, thus the results has shown that the water activity of all the three samples F1, F2 and F3 were less than 0.18. It confirms slow spoilage and higher stability of product.

7.3 Total Microbial Count

This analysis determines the total number of viable bacteria present per milliliter (ml) of the bar sample. A higher TPC indicates a greater potential for spoilage by bacteria. The method employed followed the guidelines outlined in the IS 5402:2012/ISO 4833:2003,BAM,*DGHS Manual* (2005). The highest count of microbial content is recorded upto 30×10∧1 CFU /gm.

Figure1. Total Microbial Count for Millet Bar

8. Conclusion

In conclusion, the formulation of value-added nutrition bars offers a promising approach to addressing the growing demand for convenient, healthy, and sustainable food options. Functional ingredients like popped pearl millet, black rice flakes and dates provide a boost of essential nutrients, like protein, fiber and energy. The sample F3 with 40 g of popped pearl millet, 20 g of black rice flakes, 10 g of dates and 30 g of jaggery showed best results in nutritional evaluation and was highly acceptable. the sample F3 demonstrates promising results in terms of consumer acceptance.

9. References

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